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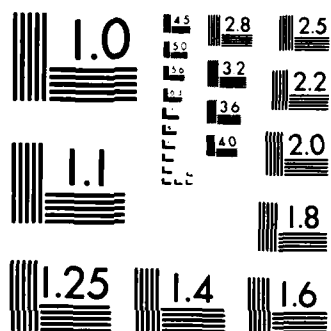
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Genetic and Physical Structure of Salmonella-coli Phage
Hybrids and Development of New Generalized
Transducing Hybrid Phages for E. Coli

Annual Report

Nobuto Yamamoto, Ph.D.

May 1984

Supported by

U.S. ARMY MEDICAL RESEARCH AND DEVELOPMENT COMMAND
Fort Detrick, Frederick, Maryland 21701-5012

Contract No. DAMD17-79-C-9134

Hahnemann University School of Medicine
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FOREWORD

Though we initially planned for development of a gene cloning vector, we have not established a recombinant DNA method for these hybrid phages during this period.

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3. Serological and genetic evidence for formation mechanis of the new tail fiber antigen of hybrids between coliphage Mu and <u>Salmonella</u> phage P22.	9
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PROGRESS

1. Intergeneric high transducing activity of $\phi 80immP22dis^-$ hybrid type

$\phi 80immP22dis^-$ hybrid type carries all the late genes of coliphage $\phi 80$ and most of the P22 early region including both the bipartite immunity region (c and Im). Such hybrids can grow in hosts lysogenic for $\phi 80immP22$ and carry not only att region but also gene 9 or a1 of P22, because these genes are situated between c and Im regions of P22. Thus the prophage of this hybrid type is inserted into the attP22 region adjacent to the proline region of the E. coli-S. typhimurium recombinant WR4027 chromosome. Induction of such a prophage creates a new hybrid type by losing the Im region and the genes between the Im and att regions of P22 and acquiring a bacterial chromosomal segment. Such new hybrids are now unable to grow in $\phi 80immP22$ lysogens, thus designated as $\phi 80immP22dis^-$. Since E. coli-S. typhimurium chromosome consists of S. typhimurium chromosomal segment coding for at least synthesis of cell wall lipopolysaccharide and E. coli segment(s) containing mal B rep, proAB and lac gene, $\phi 80immP22dis^-$ hybrids are likely to carry a coli chromosomal segment containing proline A, B and adjacent genes. Because of the lack of sufficient auxotrophic mutants in E. coli-S. typhimurium recombinant species, E. coli K12 auxotrophic mutants were used for transduction assays with $\phi 80immP22dis^-$ hybrid type. Transduction frequencies are extremely high, more than 10% for these genes. Arginine F of E. coli is a gene for ornithine carbamoyltransferase at 6min of E. coli map and is efficiently transduced by $\phi 80immP22dis^-$ hybrid type at a frequency of about 21%. Proline A of E. coli K12 was also transduced by $\phi 80immP22dis^-$ at a high frequency (about 12%) but this frequency is lower than that with arginine F. In addition methionine D was transduced with some of the

$\phi 80$ immP22dis⁻ strains but not all the $\phi 80$ immP22dis⁻ hybrids strains, whereas arginine F and proline A of E. coli K12 were transduced with all the $\phi 80$ immP22dis⁻ strains tested. These observations suggest the genetic order attP22-argF-proA-metD is in counterclockwise orientation of the E. coli chromosome.

2 The structure of the P22 homologous segment in MuimmP22 hybrids

We reported previously that MuimmP22 hybrid carries the P22 early regions including the c region and the regulatory genes of DNA synthesis (12 and 18). Infection of WR4028 with MuimmP22 produces lysogens at a high frequency. The resultant lysogens are inducible although strains lysogenic for the parent MU phage are not inducible. These results suggest that MuimmP22 hybrid carries the att and int region of P22 phage. Therefore it became desirable to determine the left end of the P22 homology in MuimmP22 hybrids.

Since WR4028 strain lysogenic for MuimmP22 is sensitive to P22 infection, superinfection of such a lysogen with P22c2ts12 induced the prophage and also produced P22 recombinants. Computation of crossovers between markers by scoring various P22 recombinant types suggests that the left arm of P22 homology in MuimmP22 hybrid ends at or near the att region of P22.

3. Serological and genetic evidence for formation mechanism of the new tail fiber antigen of hybrids between coliphage and Salmonella phage P22

MuimmP22 hybrids form plaques on smooth derivatives such as WR4028 of E. coli S. typhimurium hybrids while Mu phage infects a rough

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